#1502

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A. Title: Application for Permit for Scientific Purposes under the Endangered Species Act of 1973.

B. Species: Wenatchee River Sub-basin populations of steelhead trout (*Oncorhynchus mykiss*) and Chinook salmon (*O. tshawytscha*).

C. Date: 14 April 2004

D. Applicant Identity:

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E. Personnel, cooperators, sponsors

1. Dr. Karl Polivka will be the sole Principal Investigator responsible for compliance with permit conditions. Other PIs included Dr. Mark S. Wipfli (University of Alaska-Fairbanks), Dr. Paul A. Hessburg (PNW Research Station, USDA Forest Service), and Dr. Chris Jordan (NOAA Fisheries).

2. Field personnel: Joshua Kill, Julia Frey, 2-3 others to be determined.
3. Our study will be supported by a contract with Bonneville Power Administration, PO Box 3621, Portland 97208-3621. Final signature and approval of the contract is pending compliance with permit requirements for take of listed species that occur in the study area. Funding is expected to be available for three years, but will only be administered one year at a time. The University of Alaska at Fairbanks will be listed as a cooperator.
4. N/A
5. We have not currently made arrangements for the disposition of any dead specimens. We do not expect any individuals to be killed in this study. In the event of any mortality, we will preserve the specimen in ethyl alcohol and contact the museum at the University of Washington (Seattle WA), Box 355100, University of Washington, Seattle, WA, 98195-5100, USA.
6. N/A – we do not plan to transport or hold specimens. They will be returned alive at the location of capture.

F: Project Description, Purpose, and Significance:

1. The goals of this proposed work are to: 1) develop and test methods for monitoring headwater stream conditions at the subcatchment and stream levels, 2) determine the effects of land-use (timber harvest and roading) and vegetation cover (which is under geoclimatic control) on the biological productivity of subcatchments, and 3) use this information to relate watershed condition of fishless subcatchments to fish communities in downstream habitats. The headwaters monitoring focuses on food web productivity (i.e., the amount of aquatic arthropod biomass and organic detritus produced and exported to fish habitats) as an integrator of the processes and environmental constraints driving these ecosystems. It is also meant to determine whether food web productivity is a key determinant of the health of downstream fish communities including species of special concern listed under the Endangered Species Act of 1973.

Low-order streams (≤ 1st order; typically fishless streams) comprise more than 80% of drainage networks, yet very little is known about the role they play in affecting downstream fish habitats and communities (Benda and Dunne 1997, Gomi et al. 2002). Wipfli and Gregovich (2002) found that small fishless streams can be important energy sources for downstream food webs in salmonid ecosystems in southeastern Alaska, transporting invertebrates and organic material produced in headwaters environments to habitats lower in the drainage that contain fish. Understanding the ecological function of these headwater channels and associated subcatchments, and developing monitoring tools for assessing their condition is crucial for a broader understanding of basin-wide watershed condition, and for restoration effectiveness monitoring. Coupled with measuring selected fish community metrics (e.g., lipids, density, growth rates, behavior) in the habitats these small streams drain into, we intend to link watershed condition with stream productivity, food resources for fishes, and ultimately fish condition. Studies of the link between resource productivity and fish behavior complement studies of physical habitat features in determining the suite of habitat requirements for sustainable fish production. Social foraging theory predicts group size in a foraging habitat based on resource availability and competitive relationships (Giraldeau and Caraco 2000). Behavioral patterns in group-foraging situations can give clarity to the relationship between productivity and population health of fish. Combined studies of watershed productivity and fish condition and behavior under field conditions represent a novel approach, and could prove to be an effective means for restoration effectiveness monitoring as it applies to threatened, endangered and sensitive species in managed watersheds.

In the Interior Columbia River Basin (ICRB), many sub-basins contain anadromous and resident salmonids or evolutionarily significant units (ESUs) that have been granted status as threatened or endangered species (Interior Columbia Basin Technical Recovery Team, 2003). Status/trend and action effectiveness monitoring plans are being implemented in a number of these subbasins including the Wenatchee subbasin in central Washington State. Additionally, resident non-salmonids might be important components of aquatic food webs in the ICRB. Our approach seeks to combine current approaches in aquatic and behavioral ecology to determine the combined effect of land-use practices and climatic variation on productivity of resources relevant to the growth, condition, and behavior of listed and non-listed fishes in the ICRB at the whole-watershed scale. Changes in the biomass and taxonomic composition of the standing crop of macroinvertebrate resources, the presence of headwater-derived resources in the diets of fish and differences in fish production, population density, and foraging effort should demonstrate the contribution of headwater streams to listed and non-listed native fishes.

This work represents an opportunity to develop a novel, innovative approach for watershed assessment by directly monitoring the productivity of food webs (arguably the ultimate response of the health of stream ecosystems), and indirectly all the cumulative processes and factors (e.g., organic matter load and dynamics, inorganic sediment dynamics, flow regime, light, allochthonous inputs, nutrient and C spiraling, retention, water temperature, surficial geology, landform setting, land-use, and watershed condition) that drive it. It is important to emphasize that this work focuses on food web productivity of low order streams as a means for testing and developing a new tool for monitoring watershed condition and restoration effectiveness. This approach is novel for three reasons: (i) food web monitoring integrates the stressors, processes, and conditions that ultimately drive these ecosystems; (ii) it directly links headwater condition and downstream fish productivity; (iii) these low order watersheds comprise over 80% of typical drainage networks, therefore in aggregation have great potential to influence salmonid habitats downstream; and (iv) monitoring in these low order watersheds has been historically ignored.

2. While this project does not respond directly to a recommendation of a single federal agency, the monitoring activities, conceptual advances, and management-related information meet some of the conservation planning needs outlined by the ICRB Technical Recovery Team (2003), a cooperative body representing several state, federal and tribal agencies and charged with the mission of synthesizing and interpreting data related to the seven key ESUs in the region.

3. As described above, food web monitoring can greatly facilitate restoration efforts related to both land-use and fisheries management in watersheds in a wide variety of climates.

4. This monitoring project is conceptually linked to the Upper Columbia Basin monitoring protocol (Hillman in prep) to be implemented in the Wenatchee River sub-basin for the study of long-term variability in habitat parameters and its relationship to long-term variability in fish population sizes. We plan to augment this study of primarily physicochemical factors with a study of the links between energetic resources found in aquatic food webs and both land use and fish production.

5. We intend to study the intake of resources derived from headwater streams by all fish species encountered in the Wenatchee River sub-basin. We plan to obtain samples of macroinvertebrates consumed by all species, but we will not sacrifice listed chinook salmon and steelhead trout for lipid analysis in determining fish condition. In this scenario, we will substitute external measures of fish condition (length/mass relationships).

G. **Project Methodology**

1. This project will be conducted for a minimum of three years with an anticipated start date of 1 August 2004 and ending 30 September 2007.

2. Procedures

Small headwater subcatchments (<40 ha) will be selected in the upper Wenatchee Basin of the Columbia River Basin that consist of a fishless headwater stream that drains into streams that bear fish throughout all seasons. During some seasons, the only species present may be resident non-salmonids such as sculpins, but seasonal differences in headwater productivity might be linked to fish performance in these streams and the response of all fishes present will be considered. We expect to observe substantial variation in discharge among the headwaters. Discharge might affect the total input of invertebrates into fish-bearing systems and our statistical analyses will consider such variation. A brief description of methods includes:

- positioning sampling stations near and immediately upstream of the junctures between fishless and fish-bearing streams
- sampling with modified drift nets (Wipfli and Gregovich 2002) the biological production (invertebrates and particulate organic matter) produced in these fishless subcatchments that is delivered to fish habitats
- measuring fish density, biomass, growth rates, whole-body lipids at these fish-no fish junctions to evaluate the ecological connectedness between fishless headwaters and downstream fish performance. ***SEE "SAMPLING IN FISH-BEARING HABITAT" BELOW***
- assessing the effects of headwaters subcatchment condition on macroinvertebrate and downstream fish populations.

Sampling in Study Streams

Study streams will be small (wetted width generally <2 m). The length of stream between its origin and the sampling site will generally be less that a few kilometers. Streams selected will contain surface flow during all sampling bouts, but flow may be negligible for some streams during dry periods. Their high gradient and lack of fish habitat will likely be the factors preventing fish from colonizing reaches upstream of our sampling sites, although fish will be present downstream of study reaches. Sampling sites (points along the stream) will be selected that contain no fish, but upstream of systems with fish, to assess the actual contribution of material from fishless headwaters to fish-bearing habitats. We will confirm that our study streams lack fish by electrofishing, minnow-trapping and dip-netting reaches that plausibly could contain fish (i.e, that lack major barriers to movement such as high gradients or waterfalls).

Nutrient transport will be measured by taking two 1-L grab samples at each site every two months beginning in April for three years. Samples will immediately be placed on ice in the field and brought to the laboratory and refrigerated overnight before being express mailed to an aquatic chemistry testing facility (to be determined). Water will be tested for total phosphorus, soluble reactive phosphorus, total nitrogen, nitrate nitrogen, and ammonium nitrogen, nutrient forms that commonly limit freshwater productivity in the Pacific Northwest (Perrin et al. 1987, Johnston et al. 1997, Ashley and Slaney 1997).

Invertebrates (aquatic and terrestrial) and organic detritus (i.e., particulate organic matter ≥250-µm) will be collected with a 250-µm net attached to one end of a 75-cm long, 10-cm diameter plastic pipe frame, which will rest on the stream bottom. One frame per stream with attached net will be secured with sandbags in the middle of each stream. Because the sampler will be placed on the stream bottom, seston will be captured (suspended particulate organic matter) as well as bedload particulate organic matter, which will be collectively labeled detritus, and macroinvertebrates in the drift as well as those moving downstream along the streambed. Facilitated by high stream gradient, the downstream end of each horizontal pipe will rest above the stream surface; discharge through the sampler will be determined by recording the time taken to fill a container of known volume. Discharge will be measured during each sampling period, a mean calculated, and this value used to determine the density of invertebrates (individuals m^{-3}) and detritus (≥ 250-µm diameter, g m^{-3}). Most of the streams are expected to be sufficiently small to allow the entire streamflow to pass through the pipes. If not, the percentage relative to the total streamflow will be estimated. This fraction will be used to extrapolate the transport measured through the net for the whole stream. Replicates will be streams within each land-use and ecoregion (n = 15). Streams will be sampled continuously for invertebrates and detritus over a 48-h period once every two months annually for all sites. In addition, we will deploy drift nets in fish-bearing reaches to estimate productivity and macroinvertebrate community similarity where fish are foraging.

Invertebrates will be sorted from detritus after being placed in 70% EtOH in the field. They will be identified to the lowest reliable taxon, their body lengths measured, and dry mass determined using taxon-specific length-mass regression equations (Rogers et al. 1977; Smock 1980; Meyer 1989; Sample et al. 1993; Burgherr and Meyer 1997). Invertebrates will be categorized as either aquatic or terrestrial if they were a product of aquatic or terrestrial secondary production, respectively (Wipfli 1997). The remainder of the sample (detrital component) will be oven-dried, weighed, ashed (at 500° C for 5 h), and reweighed to determine ash-free dry mass (AFDM).

Additionally, we will measure several other physical and biological variables in the streams to link the productivity measures with causal factors in the subcatchments, including PAR, periphyton development on rock surfaces, and stream temperature, pH, and conductivity at all sites.

Sampling in Fish-Bearing Habitat

Because the macroinvertebrate community downstream of headwater junctions is likely to be affected by input of nutrients and the downstream drift of species with different taxonomic or ecological classifications than are found in the mainstem, we will sample drifting and benthic invertebrates in fish-bearing streams to determine the strength and spatial pattern of headwater stream delivery. We will deploy 4-6 drift nets of the same size used in headwater streams a minimum of 15 cm above the stream bottom for 24 hr in conjunction with the sampling of headwater streams. Benthic macroinvertebrates will also be collected with a Hess sampler at the junction of the mainstem and 50-m upstream and downstream of the junction within the mainstem to assess the effects of headwater inputs on the lower trophic levels of these fish-bearing food webs.

Fish will be captured with baited Gee minnow traps, electrofishing when necessary, and by seining in pools within 50-m downstream of the zone of contact between the fishless and fish-bearing habitats (headwater 'treatment'), and in pools within 50-m upstream of this zone (control), for determining fish responses (including diet) resulting from prey delivery from the headwater tributaries. Because we will likely encounter ESA-listed anadromous salmonids such as spring chinook salmon (*Oncorhynchus tshawytscha*) and steelhead (*O. mykiss*), we will use the most passive methods possible to estimate fish population size after obtaining the necessary permissions. Removal methods using minnow traps are much less harmful than electrofishing and impart less mechanical stress to fish than seining. With a careful sampling protocol, removal sampling with minnow traps can estimate fish density in a short (~ 1 d) time period (Bryant 2000). All captured fish will be placed in buckets of fresh stream water and monitored constantly until being returned alive to the stream. Additional estimates of fish population size will be made by snorkeling during both daytime and evening hours to account for variation in fish activity. We will conduct additional sampling of fish at pools 100 m and 150 m downstream to gain further information on the spatial extent of any effects generated by prey and nutrient input from headwater streams.

In order to link the relative contribution of invertebrate-based food webs found in the headwater streams and that of the fish-bearing streams to the condition of relevant fish populations, we must use some direct measures of fish responses to differences in food availability. Analysis of fish diet, condition and behavior will enable us to establish how strongly connected the energetic inputs of headwater streams are to the persistence of resident and anadromous species. During regular sampling of fish at our study sites we will anesthetize individuals of all species with MS-222® (50-100 mg/L unless permitting stipulations require a lower dosage) and use gastric lavage techniques (Meehan and Miller 1978) to obtain a sample of consumed prey. Although somewhat invasive, Meehan and Miller (1978) obtained high survival rates and pilot studies on hatchery-derived juvenile coho salmon (*O. kisutch*) in our study system resulted in full recovery with no immediate mortality (Polivka unpubl. data). After capture and measurement, fish will be held in buckets of fresh water, refreshed continually to maintain temperatures consistent with stream conditions, and released alive after no more than 20 min. Following sample collection and preservation, gut contents will be analyzed in the laboratory to determine whether the assemblage of taxa consumed by fish can be linked to either headwater or mainstem production. Finally, we will sacrifice 5-10 individuals of each species not listed under the ESA as permitting stipulations and local densities allow at each site for whole-body lipid analysis, a reliable indicator of fish condition (Simpkins et al. 2003). We will measure growth in 5-10 individuals of each species (chinook salmon and steelhead trout) at each site, depending on population size, by mark-and-recapture techniques. We will mark individuals > 60 mm SL with a visual implant elastomer tag (underneath the head or behind the eye) at the time of initial capture. Upon recapture, only size data will be taken prior to immediate release at the point of capture.

Foraging behavior of actively foraging drift-feeding anadromous salmonids will be quantified in the field by a single observer using focal animal surveys (Altmann 1974) in stream pools or other occupied microhabitat patches. Observations will generally not be conducted for benthic sit-and-wait foragers such as sculpins. For each fish observed, the number of foraging attempts per minute will be recorded for a thirty minute period or until the individual quits foraging in the given patch, whichever comes first. Patch residence times of shorter duration than 30 min. will be documented. Additionally, the number and identity of other individuals foraging in the patch will be recorded as will aggressive behaviors such as displays, nips, and chases between individuals. Physical dimensions of the patch will be measured and productivity estimated from drift samples described above to determine mechanistic relationships between fish production and the carrying capacity of stream reaches. Foraging theory predicts that productivity is an important determinant of group size and dynamics such as residence time at the patch scale (Giraldeau and Caraco 2000) and the use of and competition for foraging patches may be an important indicator of fish carrying capacity, particularly where headwaters may contribute to productivity.

3. Potential for injury: All capture methods involve some potential for mechanical injury of the listed species. Minnow trapping will be employed most often and will be the least injurious to the captured individuals. Seining will impose some mechanical stress, but a field technician will be present to immediately transfer fish to buckets to avoid abrasion by rocks or other materials inadvertently introduced to the seine net with fish during the course of seining. Electroshocking carries the greatest risk for injury to fish even when voltage and duration limits are strictly followed. We will only use electroshocking in microhabitats where seining and minnow trapping are impractical. Gastric lavage methods are somewhat invasive, but not lethal. Fish will be anesthetized as described above to minimize distress. Marking methods will cause a slight abrasion at the point of implant, but fish are able to recover. In all cases where fish are anesthetized, they will be allowed to recover in buckets of stream water so that they are not released into the environment while still under the influence of anesthetic. [MORTALITY ESTIMATE HERE]

H. Description and Estimates of Take

1. Identification of species:

Oncorhynchus tshawytcha, Upper Columbia River spring-run chinook salmon
Oncorhynchus mykiss, Upper Columbia River steelhead trout

2. Sampling locations and schedule

Specific locations will be determined by a partially random selection of fishless headwater streams and then sampling fish at the confluence of these headwater streams and fish-bearing habitat. We expect sampling locations in all major tributaries of the Wenatchee River sub-basin. These include the Chiwawa, Little Wenatchee, and White rivers, Nason, Chiwaukum, Peshastin, Chumstick and Mission creeks, and possibly the mainstem Wenatchee River.

The sampling schedule consists of sampling all 60 sites once every two months beginning in August 2004 and ending in August 2007. Mark-recapture, behavioral observations and gastric lavage will likely not occur at all sampling occasions. During winter months, we do not expect to encounter high densities of fish and sample size might be limiting for meaningful analyses of behavior, growth or feeding.

3. Current Status/Trend knowledge: The Upper Columbia ESU for spring chinook salmon consists of stream-type individuals that have experienced substantial declines in population due to habitat loss, hydroelectric developments, harvest pressures and impacts from supplementation programs (Interior Columbia Basin Technical Recovery Team 2003). Upper Columbia steelhead trout have declined for the same reasons. A plan has been developed by the Upper Columbia Regional Technical Team (Hillman *in prep*) to implement extensive status/trend monitoring to update the state of knowledge of population trends and correlated habitat features in the Wenatchee sub-basin. Our study is intended to complement this work by describing the effects that headwater streams on fish populations in the same study system.

4. Anticipated Take Table: All numbers very approximate and dependent on season, local conditions, etc.
Oncorhynchus tshawytcha, Upper Columbia Spring Chinook Salmon

No.	Life Stage	Take Activity	Location	Dates	Details
1800	Fry, parr, smolts	Observe	Wenatchee River subbasin	August 2004-August 2007	Total estimate for all 60 sites given (see protocol)
1500	Fry, parr	Capture, measure, weigh, release	Wenatchee River subbasin	August 2004-August 2007	Total estimate for all 60 sites given (see protocol)
600	Parr, smolts	Gastric lavage	Wenatchee River subbasin	August 2004-August 2007	Total estimate for all 60 sites given (see protocol)
600	Parr,	marking	Wenatchee	August	Total estimate

	smolts		River subbasin	2004-August 2007	for all 60 sites given (see protocol)

Oncorhynchus mykiss, Upper Columbia Steelhead Trout

No.	Life Stage	Take Activity	Location	Dates	Details
1800	Fry, parr, smolts	Observe	Wenatchee River subbasin	August 2004-August 2007	Total estimate for all 60 sites given (see protocol)
1500	Fry, parr	Capture, measure, weigh, release	Wenatchee River subbasin	August 2004-August 2007	Total estimate for all 60 sites given (see protocol)
600	Parr, smolts	Gastric lavage	Wenatchee River subbasin	August 2004-August 2007	Total estimate for all 60 sites given (see protocol)
600	Parr, smolts	Marking	Wenatchee River subbasin	August 2004-August 2007	Total estimate for all 60 sites given (see protocol)

5. There will be no intentional mortality because we do not plan to apply any of our studies that require destructive sampling to the listed species described in this proposal. We also do not expect any unintentional mortality because our techniques of capture, marking, and gastric lavage have not resulted in mortality on non-listed related species. However, it is possible that capture, marking, and gastric lavage procedures will result in mortality. We expect less than 1% mortality or fewer than 15 individuals of each species during capture based on the take tables above. Marking and gastric lavage may result in slightly higher mortality, but we expect this to be less than 2% or 12 individuals per species. Because the marking and gastric lavage activities are limited to parr and smolts, these age classes will be the only ones affected by these mortality estimates.

6. Capture rates were estimated based on previously reported calculations of population size (e.g. Hillman and Miller 2002) and snorkel/observation pilot studies conducted by this investigator. I multiplied expected encounter rates by the number of sites sampled and rounded upward to arrive at the current estimate. What we will actually capture using the methods described above may vary by season, locality, and demographic stochasticity in each population and not all of our sampling locations are expected to harbor populations of these two species. The mortality estimates provided in item (5) above are based on previous experience (0% mortality) and adjusting upward slightly to provide a maximum number. Numbers were given as a percentage to account for discrepancy between estimated and actual take.

I. Transportation and Holding

We do not plan to transport individuals from any of the study sites. All individuals will be returned alive to the stream where they were captured.

J. Breeding

Although we do not plan to maintain captive individuals for any purpose, we are willing to contribute any data and participate in any way that will be helpful to existing cooperative breeding programs.

K. Certification

"I hereby certify that the foregoing information is complete, true and correct to the best of my knowledge and belief. I understand this information is submitted for the purpose of obtaining a permit under the Endangered Species Act of 1973 (ESA) and regulations promulgated thereunder, and that any false statement may subject me to the criminal penalties of 18 U.S.C. 1001, or to penalties under the ESA."

Signature   Date

Appendix: Literature cited

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